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(54)発明の名称　高度不飽和脂肪酸成分添加人工乳

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明細書

1. 発明の名称

高度不飽和脂肪酸成分添加人工乳

2. 特許請求の範囲

1. エイコサペンタエン酸、ビスホモ-γ-リノレン酸、アラキドン酸もしくはエイコサペンタエン酸、前記脂肪酸のエステル、前記脂肪酸を含有する油脂、前記油脂の加水分解物、又は前記油脂分解物のエステル化物をそれぞれ単独で又は混合して、又は、さらにそれらにγ-リノレン酸、該脂肪酸のエステル該脂肪酸を含有する油脂、該油脂の加水分解物、又は該油脂分解物のエステル化物を加えて、添加した人工乳。

3. 発明の詳細な説明

(産業上の利用分野)

本発明は低タンパクミルク又は母乳タンパク人工乳に欠けている又は不足している高度脂肪酸成分を強化した人工乳に関する。

(従来の技術)

γ-リノレン酸、エイコサペンタエン酸、ビスホモ-γ-リノレン酸、アラキドン酸、およびエイコサペンタエン酸(各々以下のGLA、EPA、DGLA、AAA、EPAと略す)は高度不飽和脂肪酸であり、生体内では、エイコサペンタエン酸分泌調整作用等、重要な役割をもつアラキドン酸の前駆体として、これらの、二重結合を有する高度不飽和脂肪酸である。これらの、必須脂肪酸であるリノール酸やγ-リノレン酸であるジホモ-γ-リノレン酸デサチュラーゼおよびエロンガーゼと拮抗的作用酵素(Lands [illegible] 1979)として競合的である。このうちデサチュラーゼは、老化、糖尿病その他の疾病により、活性が低められ、その結果アラキドン酸やエイコサペンタエン酸の欠乏症などの種々の健康障害を引き起こすことが知られている。従って上記の高度不飽和脂肪酸を摂取することは、これらの健康障害に対する治療または予防法として有効である。

[illegible]、高度不飽和脂肪酸[illegible]ラキドン酸[illegible]を果たしていると言われ、乳児にとってこれらの[illegible]ことは、[illegible]重要な要素となっている。

[illegible]天然母乳中の[illegible]組成が[illegible]困難であることなどのため、[illegible]母乳と同等又はそれ以上の脂肪酸[illegible]であった。

[発明が解決しようとする課題]

[illegible]脂肪酸組成[illegible]天然母乳に近い脂肪酸組成を有する[illegible]を提供しようとするものである。

[課題を解決するための手段]

本発明者等は、ヒト天然母乳の[illegible]解明[illegible]脂肪酸組成を[illegible]乳[illegible]により[illegible]本発明を完成した。

従って、本発明は、[illegible]酸、[illegible]脂肪酸を含有する[illegible]脂肪酸[illegible]液体[illegible]等の人工乳を提供する。

[具体的な説明]

まず、[illegible]母乳[illegible]100 [illegible]

[illegible]脂肪酸組成を次の表に示す。

脂肪酸	母乳 (mg/ml)	牛乳 A (mg/ml)	牛乳 B (mg/ml)
[illegible]	2.0	0.9	2.3
[illegible]酸	5.4	6.2	[illegible]
[illegible]酸	1.0	0.8	0.2
[illegible]酸	2.1	3.8	1.0
[illegible]ン酸	9.4	10.2	7.5
[illegible]ル酸	1.4	5.8	5.3
[illegible]リノレン酸	0.9	0.9	0.5
[illegible]リノレン酸	0.03	tr	tr
エイコサジエン酸	0.08	0	0
ジホモ-[illegible]-リノレン酸	0.06	0	0
アラキドン酸	0.3	tr	tr
エイコサペンタエン酸	tr	tr	0
[illegible]ヘキサエン酸	tr	[illegible]	0

従って天然母乳と、これを比較した場合、人工[illegible]

[illegible]酸、アラキドン酸、[illegible]の高度不飽和脂肪酸が[illegible]で本発明においては、[illegible]人工乳[illegible]において又は製品に上記[illegible]することにより、天然母乳に[illegible]有する人工乳を得る。

この場合上記脂肪酸[illegible]異る。例えば、ヒト母乳[illegible]の経過期間と共に変化する[illegible]。脂肪酸の添加量は[illegible]する人工乳を目的と[illegible]により異る。また、人工乳の原料、人工乳の製造[illegible]によっても異る。従って、本発明においては、[illegible]て上記の脂肪酸[illegible]合わせて加える。

例えば、[illegible]酸[illegible]〜[illegible]%、エイコサジエン酸[illegible]〜[illegible]%、[illegible]リノレン酸[illegible]〜[illegible]%、アラキドン酸0.[illegible]〜0.[illegible]%、エイコサペンタエン酸[illegible]

は、本発明の製造上により製造されるが、本発明で
ることができる。例えば、上記のアセチル化
高、三重結合をもつモルホリン誘導体を原
て、発酵法、酵素法等により合成することが
ことができる。例えば、モルホリン誘導三
物を培養し、その培養液は、本発明に上記
培養、精製等を経ることにより得られる
することにより得られる脂肪酸は
を高純度で含んでおり、この脂肪
精製度として使用することができる。また、
脂肪を原料に使って加水分解することにより
酸混合物、又は脂肪酸塩混合物、例えば
な混合物が得られ、これらを本発明の脂肪酸
料として使用することができる。さらに、これら
の脂肪酸混合物を常法に従ってエステル化し、脂
肪酸エステル、例えばメチルエステル、
エステル、の混合物を得、これを本発明の脂肪
原料として使用することができる。さらに、こ
法にして得られた脂肪酸混合物又は脂肪酸
の混合物を他の脂肪酸もしくは脂肪酸エ

エステルとして精製した後、これらを使用す
ることができる。

上記の脂肪酸もしくはその塩、又は脂肪酸エス
テル、もしくはこれらの混合物は、そのまま使用
することもできるが、より良い均一性を得るため

をナイクロデキストリンの包接化合物と
一度、脱脂ミルクや全脂ミルクに添加するのが
よい。シクロデキストリンはα、β、γいずれの
タイプも用いることができる。GLA、EDA、
DGLA、ARAもしくはEPAの脂肪酸又は脂肪酸
エステルから、シクロデキストリンの包接化合物
の合成は下記のごとく行なう。シクロデキストリ
ンの結晶あるいは過飽和水溶液中に、一定量の
GLA、EDA、DGLA、ARA、EPAなどを脂肪
酸の形又は脂肪酸エステルの形で添加し、1分
ないし10時間撹拌することにより、沈殿物として
包接化合物が得られる。又、シクロデキストリン
に必要の水を加え、ミキサーで練り混ぜながら
定量のGLA、EDA、DGLA、ARA、EPA
を脂肪酸の形又は脂肪酸エステルの形で添加
1～5時間撹拌することにより包接化合物が得
られる。

本発明の人乳には、必要に応じて、
のため、トコフェロールやそのエ
ン、アスコルビン酸等、BHT
場合、0.001～0.1%、
～0.01%程度添加することが好ましい。また、
防止剤としてはこれらに限らず一般に用いられて
るものを必要に応じて使用することができる。

次に、実施例により、この発明をさらに具体的
に説明する。

実施例1

シクロデキストリンを
ルで飽和の状態に溶解し、ここにエタノール
液とEDA100mgを加え、5分間
超音波照射した。室温で約1時間、
さらに撹拌を続けながらさらに1分間超音波
射した。生成した沈殿を、遠心分離により
取し、ヘキサンで洗浄後、減圧乾燥

[illegible]混ぜ合わせ、[illegible]を調製した。

実施例2

DGLA、ARA、及び[illegible]について実施例1と同様の操作を行なった所、いずれも[illegible]を含有する[illegible]が得られた。

実施例3

EDA、DGLA、ARA、及びEPAのそれぞれのエチルエステルについて実施例1と同様の操作を行なった所、それぞれ均質なEDAエチルエステル含有ミルク、DGLAエチルエステル含有ミルク、ARAエチルエステル含有ミルク、及びEPAエチルエステル含有ミルクが得られた。

実施例4

モルティエレラ・アルピナ[illegible]（ATCC[illegible]）の培養菌体より得られた菌体油脂[illegible]石油エーテル[illegible]を用いて、[illegible]で3時間処理することによってエチルエステル化し、カラムクロマトで抽出して[illegible]

[illegible]

実施例5

[illegible]時間、[illegible]混ぜ合わせ、[illegible]を加えて1時間撹拌後、[illegible]を抽出した。[illegible]行ない、DGLA[illegible]含有[illegible]

と含む混合物を得た。この[illegible]を3種の粉末ミルク、及びSP 150[illegible]の液体ミルクに混ぜ合わせた所、いずれも均質なミルクが得られた。

実施例6

[illegible]Aエチル、EDAエチル、DGLAエチル、ARAエチル、EPAエチルをそれぞれ重量比2：1：1：1：5の割合で混ぜ合わせた混合脂肪酸エチル1gについて、実施例3と同様の操作を行なった所、均質な粉末ミルク及び液体ミルクが得られた。

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(54)-TITLE OF THE INVENTION
Foreign Language Title: Kodo fuhowa shibosan seibun tenka jinkonyu

English Title: MANUFACTURED MILK TO WHICH A HIGH-LEVEL UNSATURATED FATTY ACID COMPONENT HAS BEEN ADDED

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SPECIFICATIONS

1. Title of the Invention

Manufactured Milk To Which a High-Level Unsaturated Fatty Acid Component Has Been Added

2. Claim

1. Manufactured milk in which there has been added, either alone or in a combination, eicosadienoic acid, Bis-homo-γ-linoleic acid, arachidonic acid, or eicosapentaenoic acid, esters of the aforesaid fatty acids, oils and fats contained in the aforesaid fatty acids, or hydrolysates of the aforesaid fats and oils or an esterified product of the dissolved matter of the aforesaid fats and oils, or, in which there has been added to those materials -linolaic acid, esters of the fatty acids, oils and fats containing the fatty acids, or hydrolysates of the fatty acids or an esterified product of the dissolved matter of the fats and oils.

3. Detailed Specifications

The invention under review pertains to manufactured milk in which a minute amount of a fatty acid component, which is lacking or insufficient in manufactured milk, such as powdered milk or liquid milk, has been reinforced.

(Traditional Technology)

γ-linoleic acid, eicosadienoic acid, Bis-homo-γ-linoleic acid, arachidonic acid, and eicosapentaenoic acid (hereinafter these fatty acids are occasionally referred to as "GLA, EDA,

DGLA, ARA, and EPA") are indispensable fatty acids in sophisticated animals. In human beings, they are the starting materials in the creation of prostaglandins, which perform important functions, such as regulation of blood pressure and regulation of hormone secretion; prostaglandins are themselves high-level unsaturated fatty acids that are physiologically active. Prostaglandins are derived from linoleic acid or α-linoleic acid, which are essential fatty acids, by Δ^5-desaturase or Δ^6-desaturase and a carbon-chained elongation enzyme. The activity of the desaturases can be weakened because of aging, cancer, diabetes, and other illnesses and phenomena, and, as a result, the production of prostaglandins may be hindered. It is commonly known that if the production of prostaglandins is thwarted, that various risks to health can result. Therefore, direct intake of the aforesaid high-level unsaturated fatty acids is useful in the treatment, or in the prevention, of these health risks.

Infants obtain these high-level unsaturated fatty acids from their mothers' milk. Prostaglandins, which are derived from high-level unsaturated fatty acids, also seem to be related to a human body capability to show immunity to certain illnesses. Consequently, intake of the components for prostaglandins from their mothers' milk is certainly crucial to ensuring that newborns will enjoy healthy lives.

Nevertheless, researchers do not know for sure how much of the aforesaid fatty acids is contained in natural mothers' milk.

In addition, in that the aforesaid fatty acids are very expensive and there are other factors to consider, it is difficult to add a minute amount of fatty acids to manufactured milk, such as powdered milk or liquid milk, and it is difficult to produce powdered milk or liquid milk that contains the same quantity, or a higher quantity, of the fatty acids that are found in mothers' milk.

(Problem Points that the Invention Will Solve)

As a result of the invention under review, the minute quantity of fatty acids is made stronger, and milk that has a fatty acid component that is approximately the same as that found in human, natural mothers' milk is obtained.

(Procedures to Solve the Problem Points)

The inventors did studies on the amino acid component in human, natural milk and the fatty acid component in milk produced by conventional methods. When comparison was made, they determined whether the GLA, the DGLA, the ARA, or the EPA was sufficient in powdered milk. In addition, through another invention, the inventors had invented a method of producing these fatty acids by a method of fermentation at a low cost. As a result of these efforts, the invention under review was perfected.

Therefore, the invention under review provides manufactured milk, such as powdered milk or liquid milk, in which there has been added, either alone or in a combination, eicosadienoic acid, Bis-homo-γ-linoleic acid, arachidonic acid, or eicosapentaenoic

acid, esters of the aforesaid fatty acids, oils and fats contained in the aforesaid fatty acids, or hydrolysates of the aforesaid fats and oils or an esterified product of dissolved matter of the aforesaid fats and oils, or, in which there has been added to those materials γ-linoleic acid, esters of the fatty acids, oils and fats containing the fatty acids, or hydrolysates of the fatty acids or an esterified product of dissolved matter of the fats and oils to those materials.

(A Detailed Explanation)

The following table shows the fatty acid composition in human mothers' milk (five months after childbirth) and two types of milk (ones where a concentration of 13 g/100 ml was dissolved in water) that are available on the market.

Fatty Acids	Mothers' Milk (mg/ml)	Powdered Milk (mg/ml) A	B
Myristic acid	2.0	0.9	2.3
Palmitic acid	5.4	6.2	5.7
Palmitoleic acid	1.0	0.8	0.2
Stearic acid	2.1	2.8	1.3
Oleic acid	9.4	10.2	7.5
Linoleic acid	1.4	5.8	5.3
α-linoleic acid	0.9	0.9	0.5
γ-linoleic acid	0.03	tr	tr
Eicosadienoic acid	0.08	0	0

Bis-homo-γ-linoleic acid	0.08	0	0
Arachidonic acid	0.3	tr	tr
Eicosadienoic acid	tr	tr	0
Docosahexaenoic acid	tr	0.1	0

When comparison is made between the natural mothers' milk and the manufactured milk, it is found that the fatty acids, which are in a minute amount, such as linoleic acid, eicosadienoic acid, Bis-homo-γ-linoleic acid, arachidonic acid, and eicosapentaenoic acid, are not sufficient in the manufactured milk. Therefore, in the invention under review, fatty acids, like those indicated above, are added to the granules in the processes of making the milk or to the finished product. As a result, manufactured milk that has a fatty acid content that is approximately the same as the fatty acid content in mothers' milk will be obtained.

The amount of the aforesaid fatty acids to be added will depend upon various conditions. For example, the fatty acid composition in human mothers' milk seems to change as the time since childbirth grows longer. Therefore, the amount of fatty acids to be added will depend upon when after birth the manufactured milk is to be administered to the infant. The ingredients of the manufactured milk will also depend upon the production processes employed in the production of the manufactured milk. Therefore, in the invention under review, the aforesaid fatty acids or matter containing the fatty acids may be

added by themselves or in combinations in accordance with the conditions at hand.

For example, to a dry product may be added 0.02-0.03% γ-linoleic acid, 0.05-0.09% eicosadienoic acid, 0.05-0.08% Bis-homo-γ-linoleic acid, 0.2-0.3% arachidonic acid, or 0.01-0.03% eicosapentaenoic acid. In addition, the proper combination of matter containing complex fatty acids, for example, the utilization of lipids or hydrolysates of lipids, and a single fatty acid will strengthen the fatty acid content to the desired level. The quantity of the fatty acid or the ester of a fatty acid should be a 0.001-2 weight % for powdered milk, but it is recommended that that quantity be 0.0001-0.2% for liquid milk.

The aforesaid fatty acid can be added in a number of forms. For example, it can be added in granulated or dissolved form, or it can be added as a salt of the fatty acid, such as a sodium salt or a potassium salt. The fatty acid can also be added as an ester, such as a methyl ester or an ethyl ester. In addition, lipids that contain the aforesaid fatty acids in a high ratio, for example, triglyceride or a hydrolysate of triglyceride, or esterified products of a hydrolysate, such as esterified methyl or esterified ethyl, are examples of forms that can be utilized.

When the aforesaid fatty acids are used by themselves as individual fatty acids or two or more types of them are combined, products that have been made by acceptable methods can be put to use. For example, additives can be produced by yeast methods or fermentation methods using Mortierella microorganisms that have a

high capacity to produce the aforesaid unsaturated fatty acids. For example, after Mortierella microorganisms have been cultured and the cultured bacteria has been dried as required, it will be extracted by an organic solvent. As a result, the lipid, which is produced by evaporating, drying, and solidifying the extract, will contain the aforesaid unsaturated fatty acid in a high ratio. This lipid can be utilized as the base material for the fatty acid that pertains to the invention under review. In addition, hydrolysis of this lipid using conventional methods will produce a fatty acid compound or a fatty acid salt compound, such as a sodium salt compound. These types of compounds can then be utilized as the base material for the fatty acid that pertains to the invention under review. The esterification of these fatty acid compounds using conventional methods will produce compounds of a fatty acid ester, e.g., methyl ester or ethyl ester. These substances can then be utilized as the base material for the fatty acid that pertains to the invention under review. Similarly, after isolation of the fatty acid compounds or the fatty acid ester compounds as single fatty acids or fatty acid salts or fatty acid esters, these materials can then be utilized.

The aforesaid fatty acids or the salts of those fatty acids or fatty acid esters or compounds of them can be utilized without further processing or modification. However, so that the substances will have a higher level of consistency, it would be a good idea to add the substance to powdered milk or liquid milk

after those substances have been taken into cyclodextrin. Either an α or a ß cyclodextrin can be utilized. From a GLA, EDA, DGLA, an ARA or an EPA fatty acid or from a fatty acid ester, the syntnesis of the substance that will be taken into cyclodextrin will be as follows. GLA, EDA, DGLA, ARA, or EPA, in a specified quantity, in the form of a fatty acid or in the form of a fatty acid ester in a saturated or super-saturated aqueous solution of cyclodextrin, will be added. A substance that is taken into cyclodextrin will be produced as a deposit as a result of mixing lasting over a period of ten minutes to ten hours. In the alternative, while a small amount of water is being added to cyclodextrin and the substance is being mixed with a mixer, a specified amount of GLA, EDA, DGLA, ARA, or EPA will be added in the form of a fatty acid or in the form of a fatty acid ester. A substance that is taken into cyclodextrin will be produced as a result of mixing over a period of one to five hours.

If necessity should so dictate, tocopherol sesamol, melanoidins, a flavone derivative, or BHT may be added to the manufactured milk to prevent oxidation. If the milk is to be a powdered milk such additives should be as much as 0.0001-0.1% and if the milk is to be a liquid milk, such additives should be as much as 0.00001-0.01%. The additives mentioned are not the only additives that can be utilized as anti-oxidation agents; any such additives that are commonly known in the industry can also be used.

The examples that follow will provide a more detailed

explanation of the invention under review.

Example 1

2 g of ß-cyclodextrin is added to 20 ml of an ethanol aqueous solution. This mixture is mixed with a stirrer, and as that is occurring, 100 g of EDA is added. The substance is then incubated for two hours at 50°C. After the matter has cooled at room temperature (approximately one hour), it will be mixed again, and as that mixing is occurring, it will be incubated for ten hours at 4°C. The substance that is produced is recovered by centrifugation. After it has been rinsed in n-hexane, it will be freeze dried. As a result, 1.8 g of a substance that has been taken into cyclodextrin containing 5% EDA will be produced. 1 g of this powder will then be mixed into 1 kg of a powdered milk. As a result, a homogenized milk containing a EDA content will be produced.

Example 2

The same procedures that were used in Example 1 were repeated with DGLA, ARA, and EPA. As a result of each processing, a homogenized milk containing a DGLA content, a homogenized milk containing a ARA content, and a homogenized milk containing a EPA content were produced.

Example 3

The same procedures that were used in Example 1 were

repeated with ethyl esters of EDA, DGLA, ARA, and EPA. As a result, a homogenized milk containing a EDA ethyl ester content, a homogenized milk containing a DGLA ethyl ester content, a homogenized milk containing a ARA ethyl ester content, and a homogenized milk containing an EPA ethyl ester content were produced.

Example 4

20 g of an oil-and-fat bacteria, which was produced by a culture of Mortierella-Aeromonas SA M0219 (FERM P-8703), was esterified using an anhydride ethanol hydrochloric acid in processing lasting over a period of three hours at 50°C. The matter was then extracted in n-hexane to produce 15 g of a fatty acid ester. The composition of this substance was 16% palmitic acid ethyl, 5% stearic acid ethyl, 27% oleic acid ethyl, 10% linoleic acid ethyl, 4% GLA etnyl, 1% EDA ethyl, 7% DGLA ethyl, 20% ARA ethyl, and 10% EPA ethyl. In the same procedures as those employed in Example 1, 2 g of this fatty acid ethyl ester compound was put to use. As a result, a homogenized powdered milk was produced. In addition, when 0.1 g of the substance taken into cyclodextrin was mixed into 1 l of a liquid milk, a homogenized liquid milk was produced.

Example 5

55 ml of a 50% ethanol aqueous solution was added to 20 g of β-cyclodextrin. The substance was added to milk that had been

kept at 60°C. 2 g of DGLA ethyl was added to it, and the matter was mixed slowly for three hours. After cooling at room temperature (for two hours), the matter was incubated for ten hours at 4°C. 1 l of water was then added, and the matter was incubated for one hour. 1 l of water was added, and the matter was mixed for one hour. Thereafter, the deposit was recovered by centrifugation. The matter was then rinsed with n-hexane, and, after that, it was freeze dried. As a result, 8.5 g of a substance that had been taken into cyclodextrin containing 10% DGLA was produced. 2 g of this powder was mixed with 2 kg of a powdered milk, and 2 g of this powder was mixed with 150 l of a liquid milk. Both produced homogenized milks.

Example 6

The same procedures as those used in Example 5 were used with 2 g of fatty acid ethyl compounds into which GLA ethyl, EDA ethyl, DGLA ethyl, ARA ethyl, and EPA ethyl had been mixed in weight percentages of 2 : 1 : 6 : 4 : 8. Homogenized powdered milks and homogenized liquid milks were produced.

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